PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference PCT-7165	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/IT 03/00329	International filing date *(day/month/year)* 28.05.2003	Priority date *(day/month/year)* 31.05.2002
International Patent Classification (IPC) or both national classification and IPC C07D491/22		
Applicant SIGMA-TAU INDUSTRIE FARMACEUTICHE RIUNITE S.P.A		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 10 sheets, including this cover sheet.

   ☐ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the opinion
   - II ☐ Priority
   - III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☒ Lack of unity of invention
   - V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
10.12.2003	29.06.2004
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized Officer Weisbrod, T Telephone No. +49 89 2399-8931

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/IT 03/00329

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:

Description, Pages

1-26 as originally filed

Claims, Numbers

1-13 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

   These elements were available or furnished to this Authority in the following language: , which is:

   ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).

   ☐ the language of publication of the international application (under Rule 48.3(b)).

   ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

   ☐ contained in the international application in written form.

   ☐ filed together with the international application in computer readable form.

   ☐ furnished subsequently to this Authority in written form.

   ☐ furnished subsequently to this Authority in computer readable form.

   ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

   ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

   ☐ the description, pages:

   ☐ the claims, Nos.:

   ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

IV. Lack of unity of invention

1. In response to the invitation to restrict or pay additional fees, the applicant has:

   ☐ restricted the claims.

   ☒ paid additional fees.

   ☐ paid additional fees under protest.

   ☐ neither restricted nor paid additional fees.

2. ☐ This Authority found that the requirement of unity of invention is not complied with and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.

3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is

   ☐ complied with.

   ☐ not complied with for the following reasons:

4. Consequently, the following parts of the international application were the subject of international preliminary examination in establishing this report:

   ☒ all parts.

   ☐ the parts relating to claims Nos. .

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	2,4-6,9,10,13
	No:	Claims	1,3,7,8,11,12
Inventive step (IS)	Yes:	Claims	
	No:	Claims	1-13
Industrial applicability (IA)	Yes:	Claims	1-13
	No:	Claims	

2. Citations and explanations

   see separate sheet

Re Item I

Basis of the opinion

The application is directed to

(i) camptothecin esters (I) (claims 1-4),
(ii) process for preparing compounds (I) (independent claims 5 and 6),
(iii) compounds (I) as medicaments,
(iv) pharmaceutical compositions comprising compounds (I) (claims 8-10), and
(v) the use of compounds (I) for the preparation of a medicament (claims 11-13).

Re Item IV

Lack of unity of invention

The application as filed is considered to lack unity of invention since its subject-matter relates not to one but rather to two separate inventions not linked together by a common underlying inventive concept as required by Rules 13.1 and 13.2 PCT.

The claims and inventions to which the separate inventions relate are grouped as follows (in the order chosen by the applicant).

(1) Claims 1, 4, 7-13 (all part) and claims 2 and 6 (complete) directed to compounds (I) wherein n = m = 1, as well as subject matter referring to such compounds (I).

(2) Claims 1, 4, 7-13 (all part) and claims 3 and 5 (complete) directed to compounds (I) wherein n = m = 0, as well as subject matter referring to such compounds (I).

The identified two inventions involve the technical feature of a "camptothecin-20-O-C(O)-A- moiety" as the sole common link. However, this feature cannot be accepted to constitute a special technical feature because it does not define a contribution over the prior art. The documents D1 and D2 disclose already water soluble camptothecin prodrugs respectively derivatives comprising such "camptothecin-20-O-C(O)-A- moiety". The document D1, in particular, discloses already certain present compounds (I) wherein A is methylene, n = m = 0, and Y is $-N^+R_{12}R_{13}R_{14}$ (D1, claim 1; and examples 1 and 2) as well as their therapeutic use in treating cancer. The document D2, furthermore, teaches already "camptothecin-20-O-C(O)-A-derivatives" with a 20-substituent of the formula $-O-C(O)-(CH_2)_2-$**NH-C(O)**$-(CH_2)_m-NR^{10}R^{11}$ (cf. D2, claim 1, R^7). Certain present compounds (I) (i.e. those wherein A is C_{1-8} alkyl, n = m = 1, and Y is C_{1-8}alkyl-$NR_{12}R_{13}$; thus having a 20-substituent of the formula O-C(O)-C_{1-8}alkyl-**C(O)-NH**-C_{1-}

$_8$alkyl-$NR_{12}R_{13}$) differ from these compounds of D2 insofar as the amide group **-$(CO)_n$-$(NH)_m$-** is retroinverted in comparison with the orientation of the amide group of the compounds of the prior art.

In view of the said prior art the problem underlying the present application is seen in the provision of further water soluble derivatives of camptothecin and the contributions claimed in the present application which are possibly made over the prior art are

(a) the provision of further water soluble camptothecin derivatives by retro-inverting the orientation of the amide group of the compounds of D2; and

(b) the provision of further water soluble camptothecin derivatives by modifying e.g. the length of -A- or the nature of Y in the corresponding groups of the compounds of D1.

These contributions, however, have nothing more in common than each single of these contributions has in common with the prior art. Consequently, these contributions diverge in two different directions are, thus, not so linked as to form one single inventive concept, which would support the unity of the invention.

Re Item V

Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1 Reference is made to the following documents.

D1: US-A-4 943 579, 24.07.1990.
D2: WO 01/49691 A, 12.07.2001.
D3: Matsumoto, H. *et al. Bioorg. Med. Chem. Lett.* **2001**, *11*, 605-609; cited in the application.
D4: Crossfire Beilstein, BRN's 7967940, 7563919; & *Bioorg. Med. Chem.* **1998**, *6*, 551-562.
D5: Lerchen, H.-G.; von dem Bruch, K. *J. Prakt. Chem.* **2000**, *342*, 753-760.
D6: Crossfire Beilstein, BRN's 9312592, 9312810; & *J. Med. Chem.* **2003**, *46*, 190-193.

Document D6 was published after the priority date. Under the presumption that the priority is valid for the claimed matter the said document is not considered as

prior art under Rule 64.1 PCT.

2 Novelty

2.1 In view of D1, D4, and D5 the application does not comply with the criterion of novelty according to Article 33(2) PCT.

D1 discloses water soluble prodrugs of camptothecin according to the present formula (I) wherein R^1 to R^3 is hydrogen, A is methylene, n = m = 0, and Y is -$N^+R_{12}R_{13}R_{14}$ (D1, claim 1, R = $COCH_2NH_2$ HCl, $COCH_2NHCH_3$ HCl, $COCH_2NHC_2H_5$ HCl, $COCH_2N(C_2H_5)_2$ HCl; and examples 1 and 2), their therapeutic use in treating cancer, and the corresponding pharmaceutical formulation (claims 2 and 3). The present claims 1, 3, 7, 8, 11, and 12 lack thus novelty in view of D1.

The document, furthermore, discloses the preparation of such compounds (I) comprising reacting camptothecin with chloroacetic anhydride (i.e. an activated carboxylic acid bearing a leaving group in the omega position) to camptothecin 20-*O*-chloroacetate, reacting the chloroacetate to the corresponding iodoacetate, and substituting the iodine leaving group with a primary or secondary amine to introduce the present Y group. The process according to present claim 5 appears thus merely formally novel over D1, because according to claim 5 the camptothecin is reacted with a carboxylic acid rather than with an activated derivative thereof.

D2 relates to camptothecin-20-*O*-beta-alanine esters of improved solubility and stability. In this context the document teaches camptothecin derivatives with a substituent in position 20 of the formula -O-C(O)-$(CH_2)_2$-**NH-C(O)**-$(CH_2)_m$-$NR^{10}R^{11}$ (cf. D2, claim 1, R^7). Certain present compounds (I) (i.e. those wherein A is C_{1-8} alkyl, n = m = 1, and Y is C_{1-8}alkyl-$NR_{12}R_{13}$; thus having a 20-substituent of the formula O-C(O)-C_{1-8}alkyl-**C(O)-NH**-C_{1-8}alkyl-$NR_{12}R_{13}$) differ from these compounds of D2 insofar as the amide group -**$(CO)_n$-$(NH)_m$**- is retroinverted in comparison with the orientation of the amide group of the compounds of the prior art.

D3 relates to the design of water soluble prodrugs of sparingly water soluble drugs. The authors of the document exemplify their strategy with the prodrugs of an HIV protease inhibitor. These prodrugs comprise a prodrug moiety of the formula -O-C(O)-X-C(O)-NH-R (X is e.g. -CH_2-CH_2- and R is e.g. -$(CH_2)_2NH_2$ HCl, -$(CH_2)_2N(CH_3)_2$ HCl, or -$(CH_2)_2$COOH), which is identical to the prodrug moiety of

the present compounds (I) wherein n = m = 1. The document, however, is not relevant to the question of novelty of the present claimed matter, because the present camptothecin prodrugs (I) are not disclosed in the prior art document.

D4 and **D5** disclose certain compounds (I) as synthesis intermediates wherein n = m = 0 and Y is $-N^+R_{12}R_{13}R_{14}$ (D4, BRN's 7967940, 7563919; D5, page 754, compounds 4a-f; pages 757-759, compounds 4A-E). Present claims 1 and 3 lack thus novelty in view of these documents.

2.2 The document D6 discloses further present compounds (I) with n = m = 0 and Y = $-N^+R_{12}R_{13}R_{14}$ (BRN's 9312592, 9312810). The document may thus become relevant if the present claimed date of priority was invalid.

3 Inventive Step

3.1 The application describes the synthesis of certain compounds (I) (pages 15-26, examples 1-11) and shows that these compounds possess anticancer activity (pages 14-15). As can be understood from the application (pages 1-2, chapter "background of the invention"), these compounds (I) represent water soluble prodrugs of camptothecin, a topoisomerase I inhibitor of anticancer activity.

3.2 Invention (1) according to item IV above

D2 teaches already camptothecin derivatives as anticancer agents of improved solubility with a group -O-C(O)-$(CH_2)_2$-**NH-C(O)**-$(CH_2)_m$-$NR^{10}R^{11}$ in position 20. Certain present compounds (I) bear a group -O-C(O)-C_{1-8}alkyl-**C(O)-NH**-C_{1-8}alkyl-$NR_{12}R_{13}$ in position 20 and differ from said compounds of D2 through the orientation of the amide group of the side-chain in position 20.

Starting from **D2** as most relevant state of the art the problem underlying the aspect (1) of the application is seen in the provision of further water soluble derivatives of camptothecin.

The documents **D3** relates to the design of water soluble prodrugs of sparingly water soluble drugs by utilizing a spontaneously cleavable linker strategy. For the design of such prodrugs, two auxiliary units, a solubilizing moiety and a self-cleavable spacer, are tandemly linked to the parent drug (cf. page 605. last paragraph; and page 606, figure 2). Combinations of such solubilizing and self-

cleavable moieties are e.g. represented by the formulae -O-C(O)-$(CH_2)_2$-C(O)-NH-CH_2-COOH, -O-C(O)-$(CH_2)_2$-C(O)-NH-$(CH_2)_2NH_2$ HCl, or -O-C(O)-$(CH_2)_2$-C(O)-NH-$(CH_2)_2N(CH_3)_2$ HCl (cf. table 1) and are thus identical to the side-chain groups according to the aspect (1) of the present application. Although, the prodrug strategy of the document D3 has been exemplified with an HIV protease inhibitor, the document leaves no doubt that the general applicability of this approach is contemplated in the document D3 (cf. page 605. last paragraph; and page 606, figure 2).

As the pharmacophore of the present compounds and the compounds of D2 is represented by the camptothecin heterocycle itself (with lactone ring E being essential for cytotoxicity; the application page 1) and due to the very close structural similarity of the present compounds and the compounds of D2, it appears that the skilled person whishing to provide further camptothecin derivatives of the desired technical effect would, starting from **D2** in combination with the general teaching of **D3**, consider the present claimed compounds (I) as obvious alternatives of the compounds of D2. Consequently, in the absence of any substantiated unexpected effect of the present claimed compounds (I) in comparison with the closest related compound(s) of D2 (i.e. the present compound wherein R^1 to R^3 = H, A = -$(CH_2)_2$-, n = m = 1, and Y = -$(CH_2)_5$-NH_3^+ Cl^-; in comparison with camptothecin-20-beta-Ala-Lys ester dihydrochloride according to page 16 of D2), no inventive step would be acknowledged.

Consequently the aspect (1) according to the present claims 1, 2, 4, 6, and 7-13 does, at present, not meet the requirements of Article 33(3) PCT.

3.3 Invention (2) according to item IV above

Insofar as the aspect (2) of the application relates to novel compounds (I) the following observations would apply to the requirement of inventive step.

Certain novel compounds (I) according to the aspect (2) of the application differ from the compounds of **D1** e.g. through the length of A as representing e.g. C_{2-8} alkylene instead of methylene. Further novel compounds (I) according to the aspect (2) of the application differ from the compounds of D1 either through R^1 to R^3 or through the nature of Y.

Starting from **D1** as most relevant state of the art the problem underlying the

aspect (2) of the application is seen in the provision of further water soluble derivatives of camptothecin.

The document **D2** shows already that compounds wherein A is longer than in the compounds of **D1** (cf. D2, claim 1, R^7 = -C(O)$CH_2CH_2NR^8R^9$; i.e. A = -CH_2CH_2- instead of -CH_2-) are compatible with the desired activity, and it is even stated that β-alanine esters of camptothecin have greater stability and solubility than esters of naturally occurring amino acids (cf. page 4). Consequently, starting from D1 in combination with D2, at least certain compounds (I) of the aspect (2) of the application appear to represent merely obvious alternatives of the compounds of D1, for which no inventive step would be acknowledged unless the applicant was able to substantiate an unexpected effect in comparison with the closest related compound(s) of D1 (e.g. the present compound (I) wherein R^1-R^3 = H, n = m = 0, A = -CH_2CH_2-, Y = -$NH_3^+Cl^-$ versus example 1 of D1; or the present compound (I) wherein R^1-R^3 = H, n = m = 0, A = -CH_2CH_2-, Y = -$NHEt_2^+Cl^-$ versus example 2 of D1). Consequently the aspect (2) according to the present claims 1, 3-5 and 7-13 does, at present, not meet the requirements of Article 33(3) PCT.

In this context it is also noted that for the requirement of unity to be met the subject-matter of the aspect (2) of the application should be characterized by a common distinguishing feature over the compounds of **D1** which is at present not evident (cf. compounds (I) which differ from the compounds of D1 through A, through R^1-R^3, or through Y). Consequently, the aspect (2) of the application will, most probably, be divided into further non-unitary groups of inventions in the regional phase.

4 Deficiencies of the Application under Article 6 PCT

Claim 4 does not comply with Article 6 PCT for the following reasons.

4.1 The chemical name of example 2 (identical with entry 1 of claim 4) does not correspond with the product to be expected from the starting materials used in the preparation of example 2. This inconsistency between the experimental conditions ind the description and the compound for which protection is sought in the claim renders claim 4 unclear. In addition, the claimed compound is not comprised in the scope of claim 1 from which claim 4 is defined as dependent; this adds to the unclarity of the claim.

4.2 Furthermore, entry 3 of claim 4 (= example 4) does not fall under the scope of claim 1, thereby resulting in a lack of clarity of claim 4.

4.3 Finally, the name segments "benzylglicyl", "terbutylglycyl", and "2-methoxyphenyl-glycyl" in the entries 5-7, 9, and 10 of claim 4 leave the reader in doubt about the position of the benzyl, tert-butyl, and 2-methoxyphenyl substituents and add to the unclarity of the claim.

5 Further Deficiencies of the Application

Contrary to the requirements of Rule 5.1(a)(ii) PCT, the relevant background art disclosed in D1 and D2 is not mentioned in the description, nor are these documents identified therein.